

FIG. 1A**Nucleotide sequence of EOS (SEQ.ID.NO1)**

CCACGCGTCCGACCAGAGTCCAAGCCCTAGGCAGTGCCACCCTTACCCAGCCCAGCCTTG
AAGACAGAATGAGAGGGGTTTCCTGTCTCCAGGTCTTGCTCCTTCTGGTGCTGGGAGCTG
CTGGGACTCAGGGAAGGAAGTCTGCAGCCTGCGGGCAGCCCCGCATGTCCAGTCGGATCG
TTGGGGGCGGGGATGGCCGGGACGGAGAGTGGCCGTGGCAGGCGAGCATCCAGCATCCTG
GGGCACACGTGTGCGGGGGGTGCTCATCGCCCCCAGTGGGTGCTGACAGCGGCGCACT
GCTTCCCCAGGAGGGGCACTGCCAGCTGAGTACCGCGTGCGCCTGGGGGCGCTGCGTCTGG
GCTCCACCTCGCCCCGCACGCTCTCGGTGCCCGTGCGACGGGTGCTGCTGCCCCCGGACT
ACTCCGAGGACGGGGCCCCGCGGCGACCTGGCACTGCTGCAGCTGCGTCGCCCCGGTGCCCC
TGAGCGCTCGCGTCCAACCCGTCTGCCTGCCCGTGCCCGGCGCCCGCCCGCCCGCCGGCA
CACCATGCCGGGTACCGGCTGGGGCAGCCTCCGCCCAGGAGTGCCCCCTCCCAGAGTGGC
GACCGCTACAAGGAGTAAGGGTGCCGCTGCTGGACTCGCGCACCTGCGACGGCCTCTACC
ACGTGGGCGCGGACGTGCCCCAGGCTGAGCGCATTTGTGCTGCCTGGGAGTCTGTGTGCCG
GCTACCCCCAGGGCCACAAGGACGCCTGCCAGGGTGATTTCTGGGGGACCTCTGACCTGCC
TGCAGTCTGGGAGCTGGGTCTGGTGCGGTGAGTGGGGCAAGGGTTGTGCCCTGC
CCAACCGTCCAGGGGTCTACACCAGTGTGGCCACATATAGCCCCCTGGATTTCAGGCTCGCG
TCACTTCTAATGCTAGCCGGTGAGGCTGACCTGGAGCCAGCTGCTGGGGTCCCTCAGCCT
CCTGGTTTCATCCAGGCACCTGCCTATACCCACATCCCTTCTGCCTCGAGGCCAAGATGC
CTAAAAAAGCTAAAGGCCACCCACCCCCACCCACACCTTCTGGCTCCTCTCCTCTTT
GGGGATCACCAGCTCTGACTCCACCAACCCTCATCCAGGAATCTGCCATGAGTCCCAGGG
AGTCACACTCCCCACTCCCTTCTCTGGCTTGTATTTACTTTTCTTGGCCCTGGCCAGGGCT
GGGCGCAAGGCACGCAGTGATGGGCAAACCAATTGCTGCCCATCTGGCCTGTGTGCCCAT
CTTTTTCTGGAGAAAGTCAGATTCACAGCATGACAGAGATTTGACACCAGGGAGATCCTC
CATAGCTGGCTTTGAGGACACGGGGACCACAGCCATGAGCGGCCTCTAAGAGCTGAGAGA
CAGCCGGCAGGGAATCGGAACCCCTCAGACCCACAGCCGCAAGGCACTGGATTCTGGCAGC
ACCCTGAAGGAGCTGGGAAGTAAGTTCTTCCCCAGCCTCCAGATAAGAGCCCCGCCGGCC
AATCCCTTCATTTCAACCTAAAGAGACCCTAAGCAGAGAACCTAGCTGAGCCACTCCTGA
CCTACAAAGTTGTGACTTAATAAATGTGTGCTTTAAGCTGCCAAAAAAAAAAAA

FIG. 1B**Amino Acid sequence of EOS protease (SEQ.ID.NO.:7)**

Met Arg Gly Val Ser Cys Leu Gln Val Leu Leu Leu
 Leu Val Leu Gly Ala Ala Gly Thr Gln Gly Arg Lys
 Ser Ala Ala Cys Gly Gln Pro Arg Met Ser Ser Arg
 Ile Val Gly Gly Arg Asp Gly Arg Asp Gly Glu Trp
 Pro Trp Gln Ala Ser Ile Gln His Pro Gly Ala His
 Val Cys Gly Gly Ser Leu Ile Ala Pro Gln Trp Val
 Leu Thr Ala Ala His Cys Phe Pro Arg Arg Ala Leu
 Pro Ala Glu Tyr Arg Val Arg Leu Gly Ala Leu Arg
 Leu Gly Ser Thr Ser Pro Arg Thr Leu Ser Val Pro
 Val Arg Arg Val Leu Leu Pro Pro Asp Tyr Ser Glu
 Asp Gly Ala Arg Gly Asp Leu Ala Leu Leu Gln Leu
 Arg Arg Pro Val Pro Leu Ser Ala Arg Val Gln Pro
 Val Cys Leu Pro Val Pro Gly Ala Arg Pro Pro Pro
 Gly Thr Pro Cys Arg Val Thr Gly Trp Gly Ser Leu
 Arg Pro Gly Val Pro Leu Pro Glu Trp Arg Pro Leu
 Gln Gly Val Arg Val Pro Leu Leu Asp Ser Arg Thr
 Cys Asp Gly Leu Tyr His Val Gly Ala Asp Val Pro
 Gln Ala Glu Arg Ile Val Leu Pro Gly Ser Leu Cys
 Ala Gly Tyr Pro Gln Gly His Lys Asp Ala Cys Gln
 Gly Asp Ser Gly Gly Pro Leu Thr Cys Leu Gln Ser
 Gly Ser Trp Val Leu Val Gly Val Val Ser Trp Gly
 Lys Gly Cys Ala Leu Pro Asn Arg Pro Gly Val Tyr
 Thr Ser Val Ala Thr Tyr Ser Pro Trp Ile Gln Ala
 Arg Val Thr Ser Asn Ala Ser Arg

FIG. 2

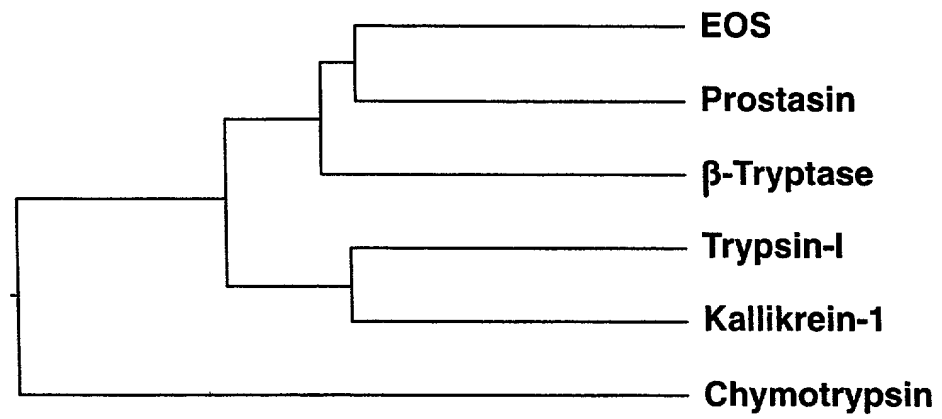


FIG. 3

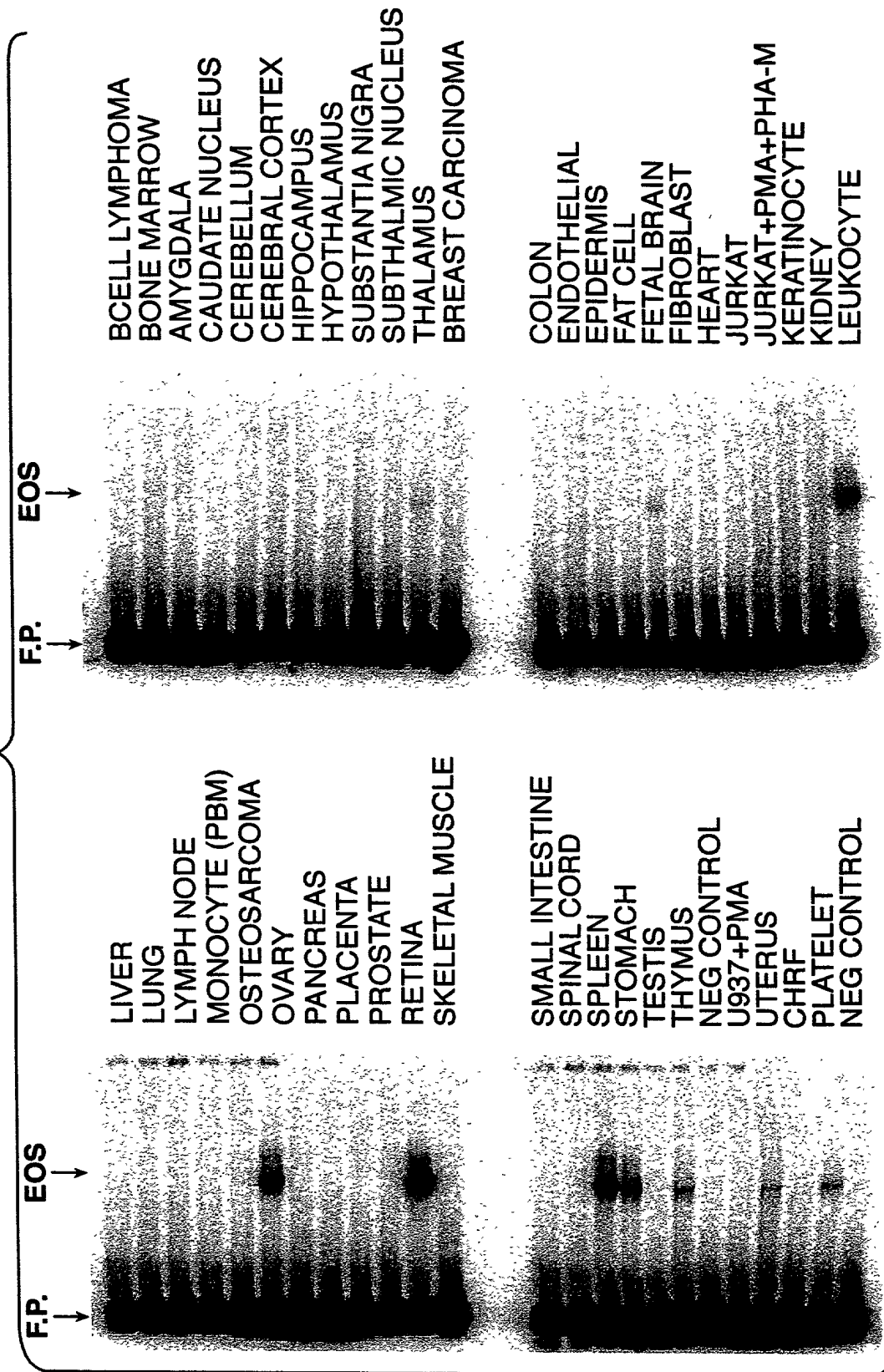


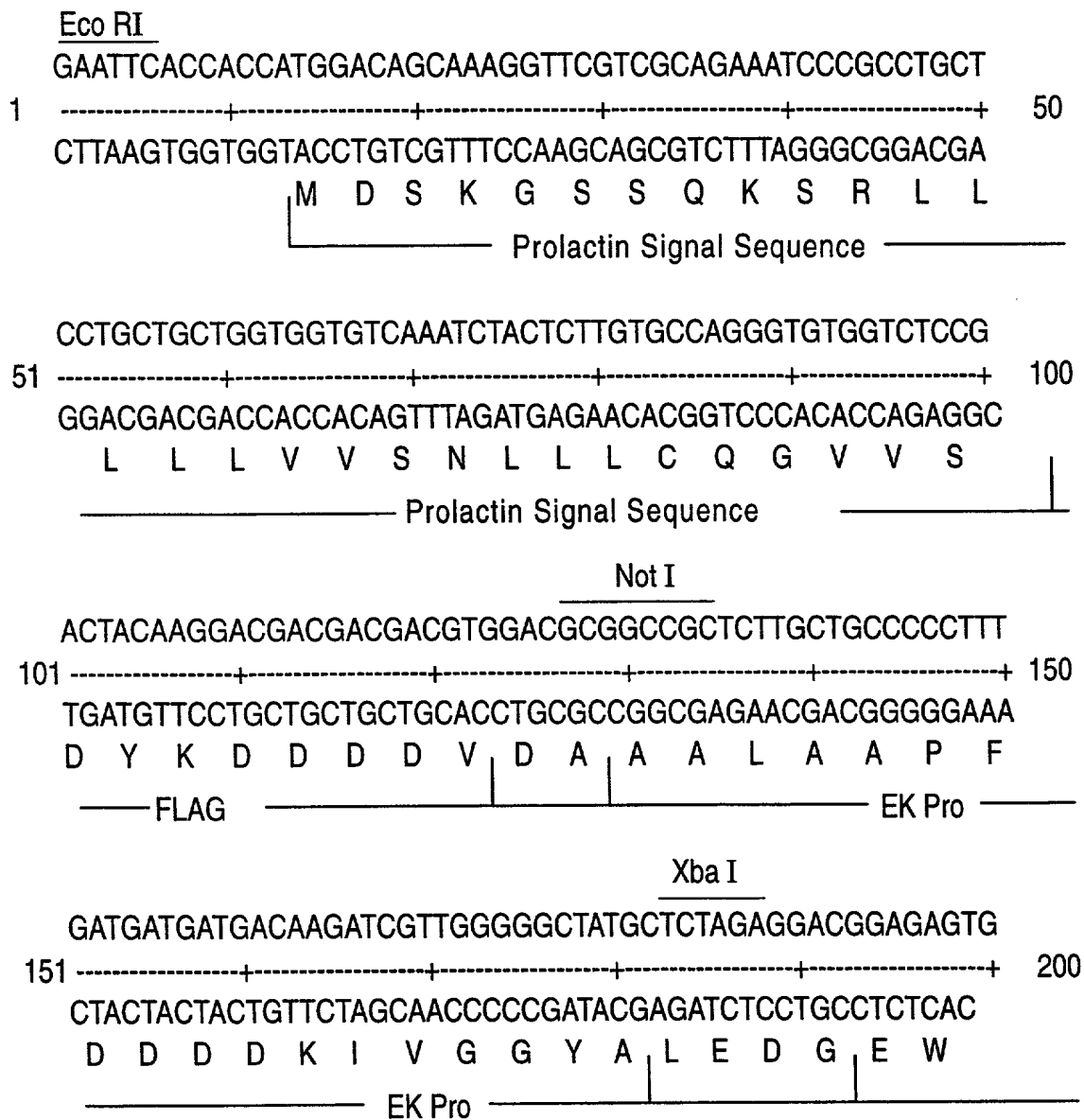
FIG. 4A

FIG. 4B

201 GCGTGGCAGGCGAGCATCCAGCATCCTGGGGCACACGTGTGCGGGGGGT
-----+-----+-----+-----+ 250
CGGCACCGTCCGCTCGTAGGTCGTAGGACCCCGTGTGCACACGCCCCCA
P W Q A S I Q H P G A H V C G G
----- Protease EOS.CDS -----

251 CGCTCATCGCCCCCAGTGGGTGCTGACAGCGGCGCACTGCTTCCCCAGG
-----+-----+-----+-----+ 300
GCGAGTAGCGGGGGGTACCCACGACTGTCGCCGCGTGACGAAGGGGTCC
S L I A P Q W V L T A A H C F P R
----- Protease EOS.CDS -----

301 AGGGCACTGCCAGCTGAGTACCGCGTGCGCCTGGGGGCGCTGCGTCTGGG
-----+-----+-----+-----+ 350
TCCCGTGACGGTCGACTCATGGCGCACGCGGACCCCGCGACGCAGACCC
R A L P A E Y R V R L G A L R L G
----- Protease EOS.CDS -----

351 CTCCACCTCGCCCCGCACGCTCTCGGTGCCCGTGCGACGGGTGCTGCTGC
-----+-----+-----+-----+ 400
GAGGTGGAGCGGGGCGTGCGAGAGCCACGGGCACGCTGCCACGACGACG
S T S P R T L S V P V R R V L L
----- Protease EOS.CDS -----

FIG. 4C

401 C C C C G G A C T A C T C C G A G G A C G G G G C C C G C G G C G A C C T G G C A C T G C T G C A G
-----+-----+-----+-----+-----+ 450
G G G G C C T G A T G A G G C T C C T G C C C G G G C G C C G C T G G A C C G T G A C G A C G T C
P P D Y S E D G A R G D L A L L Q

Protease EOS.CDS

451 C T G C G T C G C C C G G T G C C C C T G A G C G C T C G C G T C C A A C C C G T C T G C C T G C C
-----+-----+-----+-----+ 500
G A C G C A G C G G G C C A C G G G G A C T C G C G A G C G C A G G T T G G G C A G A C G G A C G G
L R R P V P L S A R V Q P V C L P

Protease EOS.CDS

501 C G T G C C C G G C G C C C G C C C G C C C G G C A C A C C A T G C C G G G T C A C C G G C T
-----+-----+-----+-----+ 550
G C A C G G G C C G C G G G C G G G C G G C G G G C C G T G T G G T A C G G C C C A G T G G C C G A
V P G A R P P P G T P C R V T G

Protease EOS.CDS

551 G G G G C A G C C T C C G C C C A G G A G T G C C C C T C C C A G A G T G G C G A C C G C T A C A A
-----+-----+-----+-----+ 600
C C C C G T C G G A G G C G G G T C C T C A C G G G G A G G G T C T C A C C G C T G G C G A T G T T
W G S L R P G V P L P E W R P L Q

Protease EOS.CDS

FIG. 4D

601 GGAGTAAGGGTGCCGCTGCTGGACTCGCGCACCTGCGACGGCCTCTACCA 650
-----+-----+-----+-----+-----+
CCTCATTCCCACGGCGACGACCTGAGCGCGTGGACGCTGCCGGAGATGGT
G V R V P L L D S R T C D G L Y H

Protease EOS.CDS

651 CGTGGGCGCGGACGTGCCCCAGGCTGAGCGCATTGTGCTGCCTGGGAGTC 700
-----+-----+-----+-----+-----+
GCACCCGCGCCTGCACGGGGTCCGACTCGCGTAACACGACGGACCCTCAG
V G A D V P Q A E R I V L P G S

Protease EOS.CDS

701 TGTGTGCCGGCTACCCCCAGGGCCACAAGGACGCCTGCCAGGGTGATTCT 750
-----+-----+-----+-----+-----+
ACACACGGCCGATGGGGGTCCCGGTGTTCTGCGGACGGTCCCACTAAGA
L C A G Y P Q G H K D A C Q G D S

Protease EOS.CDS

751 GGGGGACCTCTGACCTGCCTGCAGTCTGGGAGCTGGGTCCTGGTGGGCGT 800
-----+-----+-----+-----+-----+
CCCCCTGGGAGACTGGACGGACGTCAGACCCTCGACCCAGGACCACCCGCA
G G P L T C L Q S G S W V L V G V

Protease EOS.CDS

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FIG. 4E

801 GGTGAGCTGGGGCAAGGGTTGTGCCCTGCCCAACCGTCCAGGGGTCTACA
-----+-----+-----+-----+-----+ 850
CCACTCGACCCCGTTCCCAACACGGGACGGGTTGGCAGGTCCCCAGATGT
V S W G K G C A L P N R P G V Y

Protease EOS.CDS

851 CCAGTGTGGCCACATATAGCCCCTGGATTCAAGGCTCGCGTCACTTCTAAT
-----+-----+-----+-----+-----+ 900
GGTCACACCGGTGTATATCGGGGACCTAAGTCCGAGCGCAGTGAAGATTA
T S V A T Y S P W I Q A R V T S N

Protease EOS.CDS

Xba I
901 GCTTCTAGATACCCCTACGATGTGCCCGATTACGCCGCTAGACATCACCA
-----+-----+-----+-----+-----+ 950
CGAAGATCTATGGGGATGCTACACGGGCTAATGCGGCGATCTGTAGTGGT
A S R Y P Y D V P D Y A A R H H H

HA/HIS-TAG

Not I
951 TCACCATCACTAGCGGCCGCTTCCCTTTAGTGAGGGTTAATGCTTCGAGC
-----+-----+-----+-----+-----+ 1000
AGTGGTAGTGATCGCCGGCGAAGGGAAATCACTCCCAATTACGAAGCTCG
H H H *

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FIG. 4F

1001 -----+-----+-----+-----+ 1050
AGACATGATAAGATACATTGATGAGTTTGGACAAACCACAACCTAGAATGC
TCTGTACTATTCTATGTAACCTACTCAAACCTGTTTGGTGTGATCTTACG

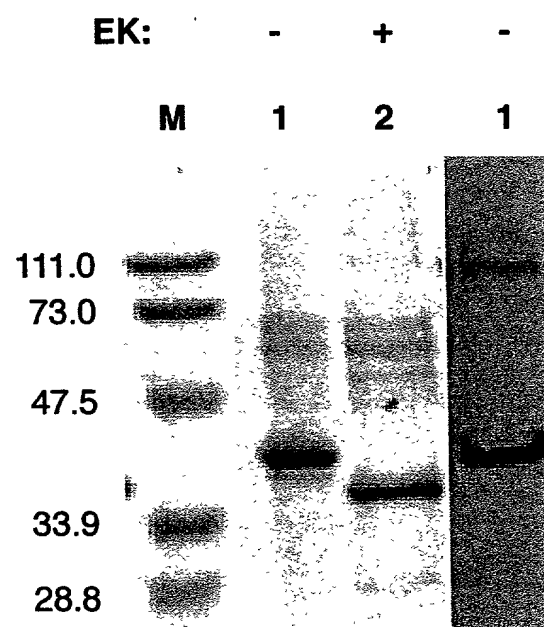
SV40 Late pA

1051 -----+-----+-----+-----+ 1100
AGTGAAAAAATGCTTTATTTGTGAAATTTGTGATGCTATTGCTTTATTT
TCACTTTTTTTACGAAATAAACACTTTAAACACTACGATAACGAAATAAA

SV40 Late pA

1101 -----+-----+ 1130
GTAACCATTATAAGCTGCAATAAACAAGTT
CATTGGTAATATTCGACGTTATTTGTTCAA

208070-16024001

FIG. 5

Protease: PFEK2-EOS-6XHIS

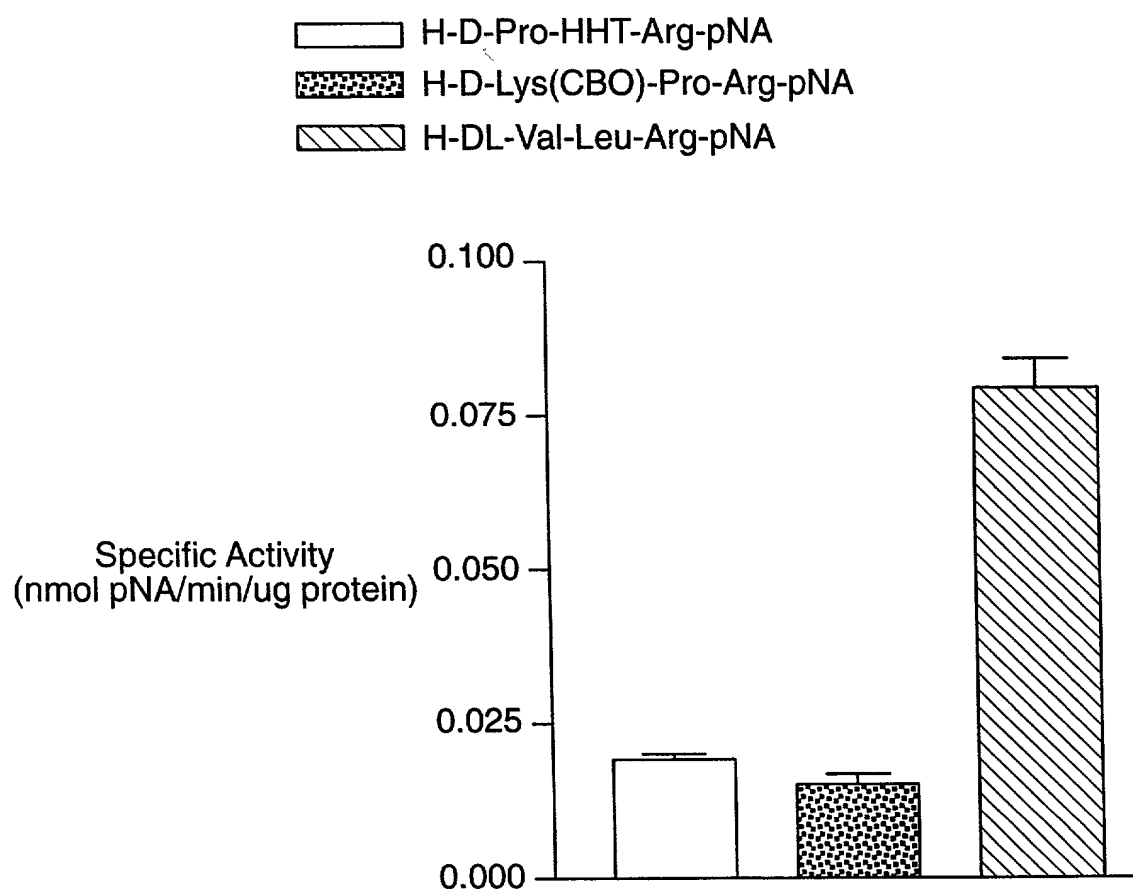
FIG. 6**Protease Activity of EOS**

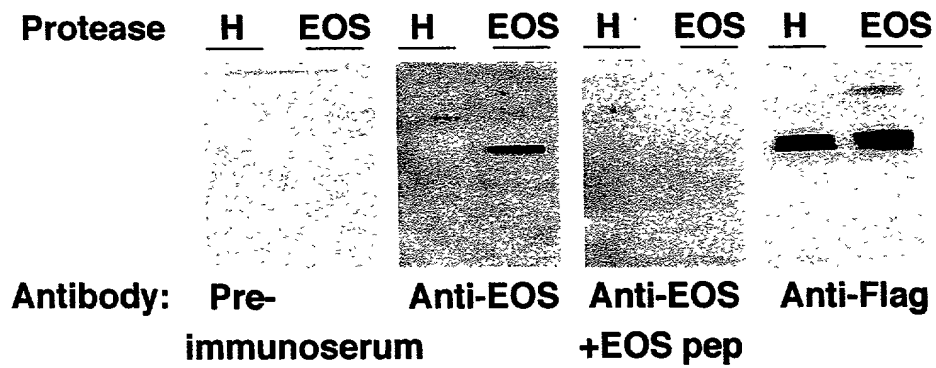
FIG. 7**Anti-EOS Antiserum Immunoblot Characterization**

FIG. 8

Localization of EOS protein (top) and mRNA (bottom)
in human spleen, lung, and colon

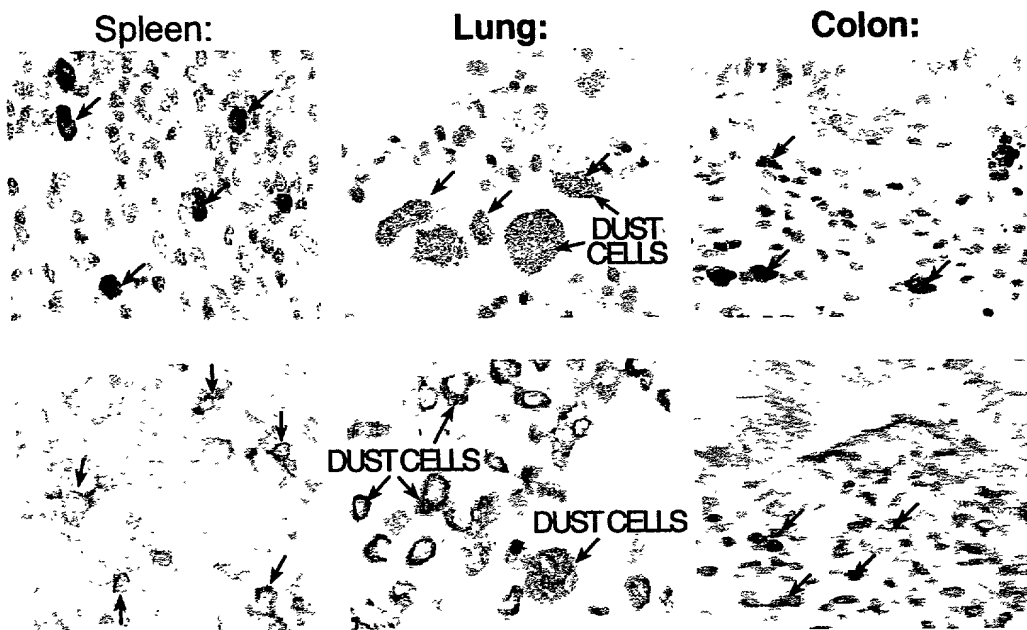
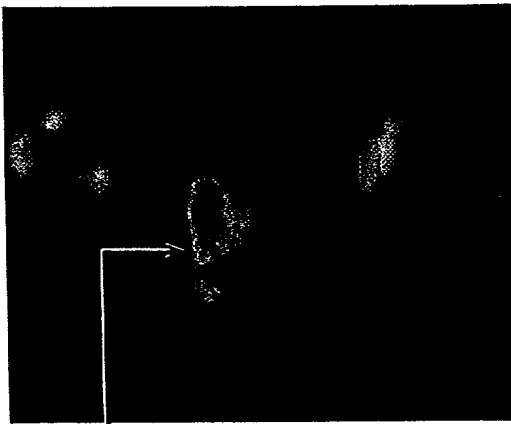
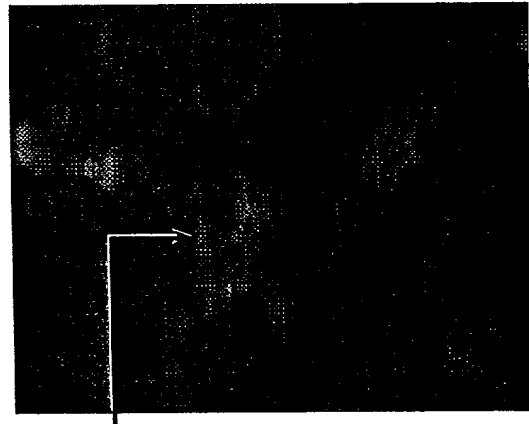


FIG. 9

Double immunofluorescence (IF:IF) of
EOS and macrophage marker:CD68



EOS in human colon



CD68 in human colon

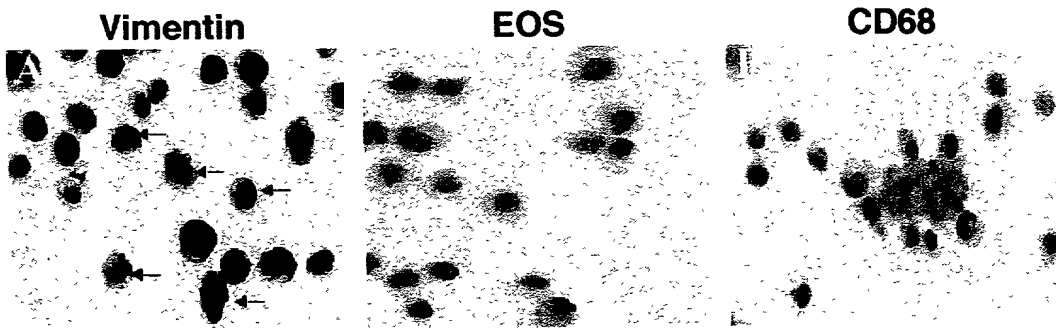
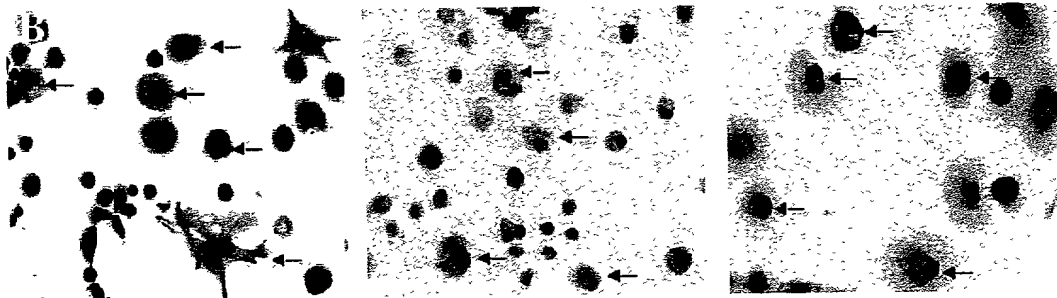
FIG. 10**Up-regulation of EOS protein by PMA in U937 cells****Untreated U937 cells:****PMA treated U937 cells:**

FIG. 11

Up-regulation of EOS mRNA by PMA in U937 cells

